

Amendments to the Claims:

The listing of claims will replace all prior versions, and listings of claims in the application:

Listing of Claims:

1. (currently amended) A recombinant, purified, or isolated polypeptide comprising an amino acid sequence selected from

(a) the sequence of SEQ ID No: 1;

(b) a sequence functionally equivalent variant of the sequence of SEQ ID NO: 1 which has greater than 77% amino acid sequence identity with SEQ ID NO: 1; and which comprises a functionally equivalent variant which is immunologically cross-reactive with and has at least substantially the same function as the original polypeptide of SEQ ID NO: 1; and

(c) a functionally equivalent fragment of a polypeptide defined in (a) or (b), wherein said functionally equivalent fragment is immunologically cross-reactive with and has at least substantially the same function as the original polypeptide of SEQ ID NO: 1.

2. (currently amended) A recombinant, purified, or isolated polypeptide comprising an amino acid sequence selected from

(a) amino acids 20 to 235 of SEQ ID NO: 1

(b) a sequence which has greater than 77% amino acid sequence identity with SEQ ID NO:1 and which comprises a functionally equivalent variant which is immunologically cross-reactive with and has at least substantially the same function as the original polypeptide of SEQ ID NO:1~~functionally equivalent variant which has greater than 77% amino acid sequence identity with amino acids 20 to 235 of SEQ ID NO: 1; and~~

(c) a functionally equivalent fragment of a polypeptide defined in (a) or (b), wherein said functionally equivalent fragment is immunologically cross-reactive with and has at least substantially the same function as the original peptide of SEQ ID NO:1.

3. (currently amended) The A-polypeptide as claimed in claim 2 wherein the sequence has greater than 90% identity with SEQ ID NO: 1.

4. (currently amended) The A-polypeptide as claimed in claim 2 wherein the sequence has greater than 99% identity with the sequence of amino acids 20 to 235 of SEQ ID NO: 1.

5. (currently amended) The A-polypeptide as claimed in claim 2 wherein the sequence is that of amino acids 20 to 235 of SEQ ID NO: 1.

6. (currently amended) The A-polypeptide as claimed in claim 1 which is obtainable from a bacterium.

7. (currently amended) The A-polypeptide as claimed in claim 1 which is obtainable from *Mycobacterium avium* subspecies *paratuberculosis*.

8. (currently amended) The A-polypeptide as claimed in claim 1 which is obtainable from a heterologous host transformed with a polynucleotide which encodes said the polypeptide comprising the sequence of SEQ ID NO:1, or a functionally equivalent variant which is immunologically cross-reactive with and has at least substantially the same function as the polypeptide of SEQ ID NO:1, or a functionally equivalent fragment thereof which is immunologically cross-reactive with and has at least substantially the same function as the polypeptide of SEQ ID NO:1 wherein said host is capable of expressing said polypeptide.

9. (currently amended) The A-polypeptide as claimed in claim 8 wherein the host is *E coli*.

10. (previously presented) A genetic construct comprising

(a) a promoter sequence;

- (b) an open reading frame polynucleotide encoding a polypeptide as claimed in claim 1;
- (c) a termination sequence.

11. (currently amended) A recombinant, purified, or isolated polynucleotide comprising the sequence of SEQ ID NO:2 or a variant thereof encoding either the polypeptide comprising the amino acid sequence of SEQ ID NO:1 or a functionally equivalent fragment thereof which is immunologically cross-reactive with and has at least substantially the same function as the polypeptide of SEQ ID NO:1 of said polynucleotide.

12. (original) A recombinant, purified or isolated polynucleotide with a nucleotide sequence complementary to the polynucleotide of claim 11.

13. (previously presented) One or more oligonucleotide or polynucleotide primers capable of amplifying a polynucleotide which encodes a polypeptide as claimed in claim 1 in a Polymerase Chain Reaction or other polynucleotide amplification method.

14. (previously presented) A purified or isolated antibody capable of binding a polypeptide as defined in claim 4 .

15. (previously presented) A vaccine composition comprising a polypeptide as claimed in claim 1 and an acceptable diluent, carrier, excipient, or adjuvant, said polypeptide being present in an amount sufficient to generate a protective immune response to *Mycobacterium avium* subspecies *paratuberculosis* infection.

16. (previously presented) A diagnostic composition for use in detecting the presence of *Mycobacterium avium* subspecies *paratuberculosis*, wherein said composition comprises a polypeptide as claimed in claim 1 .

17. (previously presented) A diagnostic composition for detecting the presence of *Mycobacterium avium* subspecies *paratuberculosis*, wherein said composition comprises a polynucleotide according to claim 11 .

18. (previously presented) A diagnostic composition for detecting the presence of *Mycobacterium avium* subspecies *paratuberculosis* comprising at least one

oligonucleotide or polynucleotide primer capable of amplifying a polynucleotide which encodes a polypeptide as claimed in claim 1 in a Polymerase Chain Reaction or other polynucleotide amplification method.

19. (original) A diagnostic composition for detecting the presence of *Mycobacterium avium* subspecies *paratuberculosis* comprising an antibody according to claim 14.

20. (previously presented) A method of detecting Johne's disease including preclinical Johne's disease in an animal comprising contacting either the animal or a sample from the animal with a polypeptide as claimed in claim 1 and detecting an immune response indicative of the presence of *Mycobacterium avium* subspecies *paratuberculosis*.

21. (currently amended) The A-method according to claim 20 wherein the response is a delayed-type hypersensitivity response.

22. (currently amended) The A-method according to claim 20 wherein said detecting comprises detecting the presence of antibodies that bind a recombinant, purified, or isolated polypeptide comprising an amino acid sequence selected from (a) amino acids 20 to 235 of SEQ ID NO: 1; (b) a functionally equivalent variant which is immunologically cross-reactive with and has at least substantially the same function as the polypeptide of SEQ ID NO:1 and which has greater than 99% amino acid sequence identity with amino acids 20 to 235 of SEQ ID NO: 1; and (c) a functionally equivalent fragment of a polypeptide defined in (a) or (b) wherein said functionally equivalent fragment is immunologically cross-reactive with and has at least substantially the same function as the original polypeptide of SEQ ID NO:1.

23. (currently amended) The A-method according to claim 22 wherein the detection of the presence of antibodies is by ELISA, radioimmunoassay or Western blotting.

24. (currently amended) A method of detecting Johne's disease including preclinical Johne's disease in an animal comprising contacting a sample from the animal either with a purified or isolated antibody capable of binding a recombinant, purified, or isolated polypeptide comprising an amino acid sequence selected from (a) amino acids 20 to 235 of SEQ ID NO: 1, (b) a functionally equivalent variant which is immunologically cross-

reactive with and has at least substantially the same function as the original protein and
which has greater than 99% amino acid sequence identity with amino acids 20 to 235 of
SEQ ID NO: 1, and (c) a functionally equivalent fragment of a polypeptide defined in (a)
or (b) wherein said functionally equivalent fragment is immunologically cross-reactive
with and has at least substantially the same function as the original polypeptide of SEQ
ID NO:1; or a composition comprising an antibody specific to the recombinant, purified,
or isolated polypeptide comprising an amino acid sequence selected from (a) amino acids
20 to 235 of SEQ ID NO: 1, (b) a functionally equivalent variant which is
immunologically cross-reactive with and has at least substantially the same function as
the polypeptide of SEQ ID NO:1 which has greater than 99% amino acid sequence
identity with amino acids 20 to 235 of SEQ ID NO: 1, and (c) a functionally equivalent
fragment of a polypeptide defined in (a) or (b) and wherein said functionally equivalent
fragment is immunologically cross-reactive with and has at least substantially the same
function as the original polypeptide of SEQ ID NO:1; and detecting a polypeptide which
binds to the antibody.

25. (currently amended) The A-method according to claim 24 wherein the presence
of bound antibody is determined by ELISA, radioimmunoassay or Western blotting.

26. (currently amended) The A-method according to claim 24 for detecting the
presence of *Mycobacterium avium* subspecies *paratuberculosis* at a preclinical phase of
Johne's disease.

27. (previously presented) A method of detecting Johne's disease including preclinical
Johne's disease in an animal comprising contacting a sample from the animal with a
composition comprising of at least one oligonucleotide or polynucleotide primers capable
of amplifying a polynucleotide which encodes a polypeptide as claimed in claim 4 in a
polynucleotide amplification method and detecting the amplification product.

28. (currently amended) The A-method as claimed in claim 27 wherein the
polynucleotide amplification method is a polymerase chain reaction method.

29. (currently amended) The ~~A~~—method according to claim 22 for detecting the presence of *Mycobacterium avium* subspecies *paratuberculosis* at a preclinical phase of Johne's disease.

30. (previously presented) A method of detecting Johne's disease in an animal comprising contacting a sample from the animal with a composition comprising a polynucleotide capable of binding to a polynucleotide which encodes a polypeptide as claimed in claim 4 .

31. (currently amended) The ~~A~~—method according to claim 30 wherein said polynucleotide is detectably labeled.

32. (currently amended) The ~~A~~—method according to claim 31 wherein said detectable label is a radioisotope or fluorescent tag.

33. (previously presented) A method of prophylactically or therapeutically treating an animal against Johne's disease which comprises administering to an animal a polypeptide as claimed in claim 1 to produce a protective immunological response in the animal.

34. (currently amended) The ~~A~~—method according to claim 33 which is a therapeutic method.

35. (currently amended) The ~~A~~—method according to claim 33 which is a prophylactic method.

36. (original) A method of vaccinating against Johne's disease which comprises administering to an animal a vaccine composition as claimed in claim 15 in an amount sufficient to produce a protective response.

37. (currently amended) The ~~A~~—method according to claim 36 wherein said administration is performed on a single occasion.

38. (currently amended) The ~~A~~—method according to claim 36 wherein said administration is performed on more than one occasion.

39. (currently amended) The A method as claimed in claim 36 ~~3~~ wherein ~~0.1-100011G/Kg~~ 0.1-1000μg/Kg is administered of a recombinant, purified, or isolated polypeptide comprising an amino acid sequence selected from

(a) the sequence of SEQ ID ~~NO:1~~ NO:1;

(b) a ~~functionally equivalent variant of the sequence of SEQ ID NO:1~~ which has greater than 77% amino acid sequence identity with SEQ ID NO:1 and which comprises a functionally equivalent variant which is immunologically cross-reactive with and has at least substantially the same function as the original polypeptide of SEQ ID NO:1; and

(c) a functionally equivalent fragment of a polypeptide defined in (a) or (b) wherein said functionally equivalent fragment is immunologically cross-reactive with and has at least substantially the same function as the original polypeptide of SEQ ID NO:1.

40. (currently amended) The A method as claimed in claim 39 wherein ~~5-500μG/Kg~~ 5-500μg/Kg of the polypeptide is administered.

41. (previously presented) A kit for use in detecting the presence of *Mycobacterium avium* subspecies *paratuberculosis* comprising at least two of the following:

a polypeptide as claimed claim 1 ;

an antibody that binds said polypeptide, and

a reagent for determining antigen-antibody binding.

42. (previously presented) A host cell transformed with a polynucleotide of claim 11.

43. (original) A vector comprising the construct as claimed in claim 10.

44. (original) A host cell incorporating a construct of claim 10.

45. (original) A host cell incorporating a vector as claimed in claim 43.

46. (currently amended) The A host cell according to claim 45 wherein said vector exists within the host cell as a plasmid.

47. (currently amended) The A-host cell according to claim 45 wherein said vector is integrated into the genome of the host cell.

48. (currently amended) The A-method as claimed in claim 20 wherein the animal is a ruminant.

49. (currently amended) The A-method as claimed in claim 47 wherein the animal is a sheep.

50. (currently amended) The A-method as claimed in claim 33 wherein the animal is a ruminant.

51. (currently amended) The A-method as claimed in claim 50 wherein the ruminant is a sheep.